(FILE 'HOME' ENTERED AT 17:50:10 ON 28 OCT 2002)

FILE 'BIOSIS, CABA, CAPLUS, EMBASE, LIFESCI, MEDLINE, SCISEARCH, USPATFULL, JAPIO' ENTERED AT 17:51:10 ON 28 OCT 2002

FILE 'AGRICOLA, LIFESCI, CONFSCI, BIOSIS, VETU, VETB, PHIN, PHIC' ENTERED AT 17:51:18 ON 28 OCT 2002

| | | | |
|---|---|---|---|
| L1 | 3352 | S | CRYPTOSPORIDIUM PARVUM |
| L2 | 776 | S | L1 AND (BOVINE OR CALF OR CALVES OR COW OR COWS OR CATTLE) |
| L3 | 0 | S | L1 AND P21 |
| L4 | 0 | S | L1 AND P21 ANTIGEN |
| L5 | 34 | S | CP15 |
| L6 | 24 | S | L1 AND L5 |
| L7 | 11 | DUP REM | L6 (13 DUPLICATES REMOVED) |
| L8 | 17 | S | P21 ANTIGEN |
| L9 | 0 | S | L1 AND L8 |
| L10 | 16 | DUP REM | L8 (1 DUPLICATE REMOVED) |

7 ANSWER 1 OF 11 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.

AB Preparturient cows were immunized three times over a six-week period with recombinant plasmid DNA encoding the **Cryptosporidium parvum CP15**/60 antigen by injecting the DNA in the mammary gland. Serum was collected at each immunization and first colostrum was collected after parturition; all were assayed for Cryptosporidium-specific antibodies (Ab). A serological response to C. parvum sporozoite and oocyst antigen was detected in cows immunized with pCP15/60 plasmid DNA. Colostrum from these cows, unlike colostrum from normal controls, contained Ab specific for C. parvum sporozoites and oocysts as indicated by immunofluorescence Ab (IFA) staining. Colostrum was also tested for conferring passive immunity against C. parvum infection by oral administration to immunosuppressed adult inbred mice. Immune colostrum and control colostrum were administered to separate groups of dexamethasone (DEX)-treated adult C57BL/6NCr mice beginning 12 h before and at 12 h intervals for 3 days afteroral C. parvum oocyst infection. Cryptosporidium development was assayed in ilea of immune- and control-colostrum-treated mice 96 h postinfection by semiquantitative PCR. Mice receiving immune colostrum showed partial protection (about 50% reduction) against intestinal C. parvum development compared to mice receiving control colostrum. This protection was evident at a challenge dose of 103 C. parvum oocysts per mouse; no differences were noted in parasite development between groups receiving immune or control colostrum and infected with 104 oocysts. This study showed that serum and colostrum Ab response to C. parvum can be elicited in preparturient cows by direct injection of recombinant pCP15/60 plasmid DNA and that passive protection against cryptosporidiosis can be obtained by treating immunosuppressed mice with immune colostrum before and after C. parvum infection.

AN 1999:351263 BIOSIS

DN PREV199900351263

TI Hyperimmune bovine colostrum specific for recombinant **Cryptosporidium parvum** antigen confers partial protection against cryptosporidiosis in immunosuppressed adult mice.

AU Jenkins, M. C. (1); O'Brien, C.; Trout, J.; Guidry, A.; Fayer, R.

CS (1) Immunology and Disease Resistance Laboratory, Agricultural Research Service, USDA, Beltsville, MD, 20705 USA

SO Vaccine, (May 14, 1999) Vol. 17, No. 19, pp. 2453-2460. ISSN: 0264-410X.

DT Article

LA English

SL English

L7 ANSWER 2 OF 11 LIFESCI COPYRIGHT 2002 CSA DUPLICATE 1

AB In this study the effectiveness of a DNA vaccine to confer protection against cryptosporidiosis, an enteric infection of lifestock and humans, was evaluated. A vaccination protocol using a recombinant plasmid encoding the 15 kDa surface sporozoite protein of **Cryptosporidium parvum** was developed in adult pregnant goats. The present study reports that nasal immunization of pregnant goats with **CP15**-DNA led to a transfer of immunity to offspring conferring protection against C. parvum infection. Kids from **CP15**-DNA-vaccinated dams shed significantly fewer oocysts and over a shorter period than did kids from unvaccinated goats. The low level of parasite development in protected kids did not affect their growth whereas unprotected kids grew much slowly. There was still a significant difference in the weights of protected and unprotected kids after complete recovery. Anti-**CP15** antibodies were present in serum and colostrum from vaccinated goats. Nevertheless, the precise immune mechanism of protection has still to be determined. This vaccine should reduce the economic losses due to cryptosporidiosis in ruminants, specially in small ruminants (calves, lambs, kids). It has also the potential to reduce environmental contamination by reducing oocyst shedding.

AN 1999:83281 LIFESCI

TI Protection of kids against **Cryptosporidium parvum**

infection after immunization of dams with **CP15**-DNA
AU Sagodira, S.; Buzoni-Gatel, D.; Iochmann, S.; Naciri, M.; Bout, D.
CS Equipe Associee INRA d'Immunologie Parasitaire, UFR des Sciences Pharmaceutiques, 31 Avenue Monge, 37200 Tours, France; E-mail: sagodira@univ-tours.fr
SO Vaccine, (19990514) vol. 17, no. 19, pp. 2346-2355.
ISSN: 0264-410X.
DT Journal
FS F; K
LA English
SL English

L7 ANSWER 3 OF 11 LIFESCI COPYRIGHT 2002 CSA DUPLICATE 2
AB DNA immunization offers a novel approach to inducing humoral and cellular immunity against infectious pathogens. We examined whether such an approach could be used against cryptosporiodiosis, an intestinal disease caused by the protozoan parasite **Cryptosporidium parvum** . This infection is a major problem for young ruminants and immunosuppressed individuals in whom cryptosporidiosis causes life-threatening symptoms. The life cycle of C. parvum takes place in the enterocytes of the intestinal epithelium. We therefore focused our attention on a route of immunization that might induce a mucosal immunoglobulin (Ig)A response. Eight-week-old BALB/c mice were immunized intranasally with DNA encoding a 15-kDa C. parvum sporozoite antigen ( **CP15**-DNA) cloned onto the plasmid pcDNA3. **CP15** -DNA-immunized mice developed specific and longlasting production of anti-**CP15** Ig A in intestinal secretions and specific IgG in sera 3 months and 1 year after the first DNA inoculation. **CP15** -DNA-immunized mice also developed an antigen-specific T lymphocyte proliferative response in both spleen and mesenteric lymph nodes. Control mice that received the pcDNA3 plasmid alone did not develop specific humoral and cellular responses. These results indicate that plasmid DNA may provide a powerful means of eliciting intestinal humoral and cellular responses to C. parvum infections in mammals.
AN 2000:22058 LIFESCI
TI Nasal immunization of mice with **Cryptosporidium parvum** DNA induces systemic and intestinal immune responses
AU Sagodira, S.; Iochmann, S.; Mevelec, M.-N.; Dimier-Poisson, I.; Bout, D.
CS Equipe Associee INRA d'Immunologie Parasitaire, UFR des Sciences Pharmaceutiques, 31 Avenue Monge, 37200 Tours, France
SO Parasite Immunology [Parasite Immunol.], (19991000) vol. 21, no. 10, pp. 507-516.
ISSN: 0141-9838.
DT Journal
FS F; K
LA English
SL English

L7 ANSWER 4 OF 11 AGRICOLA DUPLICATE 3
AN 1999:59717 AGRICOLA
DN IND21998858
TI Comparison of the humoral and cellular immune responses to two preparations of **Cryptosporidium parvum CP15** /60 recombinant protein.
AU Iochmann, S.; Sagodira, S.; Mevelec, M.N.; Reperant, J.M.; Naciri, M.; Coursaget, P.; Bout, D.
CS Equipe INRA d'Immunologie Parasitaire, Nouzilly, France.
AV DNAL (QR175.M53)
SO Microbial pathogenesis, June 1999. Vol. 26, No. 6. p. 307-315
Publisher: London ; Orlando : Academic Press, c1986-
CODEN: MIPAEV; ISSN: 0882-4010
NTE Includes references
CY England; United Kingdom
DT Article

FS Non-U.S. Imprint other than FAO
LA English

L7 ANSWER 5 OF 11 LIFESCI COPYRIGHT 2002 CSA DUPLICATE 4
AB An improved semiquantitative technique was developed for measuring low infectious doses of **Cryptosporidium parvum** in neonatal mice using polymerase chain reaction (PCR). Separate litters of neonatal mice were inoculated with 0, 10 super(2), 10 super(3), or 10 super(4) C. parvum oocysts and killed 96 hr postinfection. A segment of the ileum or the entire whole intestine was then removed from subgroups of mice in each litter and total DNA was extracted using standard procedures. By employing a **CP15**/60-based semiquantitative PCR technique, C. parvum DNA was detected in mice infected with as few as 10 super(2) oocysts. DNA isolated from the ileum of infected mice produced a more intense PCR signal than DNA isolated from the whole intestine. This technique was used to study the intracellular development of C. parvum sporozoites that had been exposed in the oocyst stage to either 0, 15, 20, 25, or 30 kRad gamma -irradiation. A **CP15**/60 PCR signal was observed in ileum tissue from mice infected with 0-kRad- or 15-kRad-irradiated C. parvum oocysts. A very slight PCR signal was generated by PCR on ileum tissue DNA from mice infected with 20-kRad-irradiated oocysts, whereas no signal was observed in PCR on intestinal DNA from mice infected with oocysts exposed to higher radiation doses.
AN 1999:728 LIFESCI
TI Development and application of an improved semiquantitative technique for detecting low-level **Cryptosporidium parvum** infections in mouse tissue using polymerase chain reaction
AU Jenkins, M.C.; Trout, J.; Fayer, R.
CS Immun. and Dis. Resistance Lab., Agric. Res. Serv., USDA, Bldg. 1040, BARC-EAST, Beltsville, MD 20705, USA
SO J. PARASITOL., (19980200) vol. 84, no. 1, pp. 182-186. ISSN: 0022-3395.
DT Journal
FS K
LA English
SL English

L7 ANSWER 6 OF 11 LIFESCI COPYRIGHT 2002 CSA DUPLICATE 5
AB A semi-quantitative method for measuring **Cryptosporidium parvum** infection in mice using polymerase chain reaction (PCR) was developed for evaluating therapeutic reagents against cryptosporidiosis. A competitor molecule composed of **CP15**/60 primer-binding sequences flanking an unrelated sequence was synthesized and used in PCR to amplify competitor and target **CP15**/60 sequences. For estimating relative intestinal parasite numbers, seven-day-old BALB/C mice were infected with 0, 102, 103, or 104 C. parvum oocysts. At 24, 48, 72, and 96 h post-infection, three mice per group were killed and the intestine from each mouse was processed for DNA. Semi-quantitative PCR using serial dilutions of competitor molecule and a constant amount of mouse tissue DNA showed that challenge doses of 103 and 104 oocysts could be easily detected at both 72 and 96 h post-infection. **CP15**/60-specific PCR products were not observed at earlier timepoints (24 and 48 h) nor at any timepoint with the 0 or 102 challenge doses. The semi-quantitative PCR technique proved to be more sensitive, rapid, and cost-effective compared to conventional histological scoring of cryptosporidial stages in tissue sections from C. parvum-infected mice. A rapid method for extracting DNA from infected mouse intestine was developed and, when used in the semi-quantitative PCR, proved to be as reproducible as conventional DNA extraction methods.
AN 97:96947 LIFESCI
TI A semi-quantitative method for measuring **Cryptosporidium parvum** infection using polymerase chain reaction
AU Jenkins, M.; Trout, J.; Fayer, R.
CS Immunology and Disease Resistance Laboratory, Agricultural Research

Service, USDA, BARC-EAST, Building 1040 Beltsville, MD 20705 USA
SO J. MICROBIOL. METHODS, (1997) vol. 28, no. 2, pp. 99-107.
ISSN: 0167-7012.
DT Journal
FS K
LA English
SL English

L7 ANSWER 7 OF 11 LIFESCI COPYRIGHT 2002 CSA DUPLICATE 6
AB In an effort to generate high titer colostrum for immunotherapy of cryptosporidiosis, a study was conducted to test the efficacy of immunizing sheep with recombinant plasmid DNA (pCMV-**CP15**/60) encoding epitopes of 15 and 60 kDa surface antigens of **Cryptosporidium parvum** sporozoites. The plasmid DNA was used to immunize preparturient ewes at three dose levels by jet-injection into either hind limb muscle (IM) or mammary tissue (IMAM). Regardless of route of injection, a dose-dependent anti-**CP15**/60 immunoglobulin response was observed in sera and colostrum from sheep immunized with pCMV-**CP15**/60 plasmid DNA. High titer antibody responses were observed in one of three animals per group receiving an IM injection of 100 or 1000 mu g pCMV-**CP15**/60. IMAM immunization with 100 or 1000 mu g pCMV-**CP15**/60 plasmid DNA elicited higher titer colostrum responses and more consistent serum responses compared to IM injections. A negligible serum and colostrum anti-**CP15**/60 response was observed in ewes injected IM with 10 mu g pCMV-**CP15** /60 or 1000 mu g control plasmid DNA. Immunoblotting of native C. parvum sporozoite/oocyst protein with hyperimmune serum and colostrum corroborated the increased titers against **CP15**/60 antigen. Serum and colostrum antibodies from pCMV-**CP15**/60-immunized sheep were eluted from native **CP15** protein and bound a surface antigen of C. parvum sporozoites as indicated by indirect immunofluorescence staining.
AN 96:27513 LIFESCI
TI Serum and colostrum antibody responses induced by jet-injection of sheep with DNA encoding a **Cryptosporidium parvum** antigen
AU Jenkins, M.; Kerr, D.; Fayer, R.; Wall, R.
CS Parasite Immunobiol. Lab., LPSI, Agric. Res. Serv., USDA, Beltsville, MD 20705, USA
SO VACCINE, (1995) vol. 13, no. 17, pp. 1658-1664.
ISSN: 0264-410X.
DT Journal
FS F; K
LA English
SL English

L7 ANSWER 8 OF 11 AGRICOLA DUPLICATE 7
AN 96:17275 AGRICOLA
DN IND20502850
TI Cloning and expression of cDNA encoding an antigenic **Cryptosporidium parvum** protein.
AU Jenkins, M.C.; Fayer, R.
CS Parasite Immunobiology Laboratory, ARS, USDA, Beltsville, MD.
AV DNAL (QL757.M6)
SO Molecular and biochemical parasitology, Apr 1995. Vol. 71, No. 1. p. 149-152
Publisher: Amsterdam : Elsevier Science Publishers, B.V.
CODEN: MBIPDP; ISSN: 0166-6851
NTE Includes references
CY Netherlands
DT Article
FS Non-U.S. Imprint other than FAO
LA English

L7 ANSWER 9 OF 11 LIFESCI COPYRIGHT 2002 CSA DUPLICATE 8

AB Sporozoites of **Cryptosporidium parvum** were examined after gliding upon glass microscope slides using monoclonal antibodies to the 15 and 25 kDa surface molecules and immunogold-silver enhancement. Both antibodies bound to surface antigen deposited as trails behind parasites, suggesting that both surface molecules are involved in substrate attachment.
AN 95:16354 LIFESCI
TI Both **CP15** and CP25 are left as trails behind gliding sporozoites of **Cryptosporidium parvum** (Apicomplexa)
AU Tilley, M.; Upton, S.J.*
CS Dep. Clin. Sci., London Sch. Hyg. Trop. Med., Keppel St., London WC1E 7HT, UK
SO FEMS MICROBIOL. LETT., (1994) vol. 120, no. 3, pp. 275-278. ISSN: 0378-1097.
DT Journal
FS K
LA English
SL English

L7 ANSWER 10 OF 11 AGRICOLA DUPLICATE 9
AB A cDNA (**CP15**/60) encoding epitopes of **Cryptosporidium parvum** 15- and 60-kDa sporozoite proteins was isolated and expressed in Escherichia coli toward the goal of developing an immunogen for producing high-titer anticryptosporidial colostrum. Antisera prepared in rats to native C. parvum 15-kDa protein and used to identify the **CP15**/60 bacteriophage clone recognized both 15- and 60-kDa in vitro translation products derived from sporozoite RNA. Antisera specific for recombinant **CP15**/60 antigen recognized native 15- and 60-kDa C. parvum sporozoite proteins by immunoblotting and identified both surface and internal antigens on C. parvum sporozoites by immunofluorescence staining. Northern (RNA) and Southern blot hybridization experiments using sporozoite RNA and DNA indicated that **CP15**/60 DNA is transcribed as a single 1.4-kb RNA species from a single-copy gene. Recombinant **CP15**/60 antigen was recognized by hyperimmune colostrum from cows immunized with C. parvum oocyst-sporozoite protein and by convalescent-phase sera from C. parvum-infected calves.
AN 93:84479 AGRICOLA
DN IND20337157
TI Cloning and expression of a cDNA encoding epitopes shared by 15- and 60-kilodalton proteins of **Cryptosporidium parvum** sporozoites.
AU Jenkins, M.C.; Fayer, R.; Tilley, M.; Upton, S.J.
AV DNAL (QR1.I57)
SO Infection and immunity, June 1993. Vol. 61, No. 6. p. 2377-2382 Publisher: Washington, D.C., American Society for Microbiology ISSN: 0019-9567
NTE Includes references
CY District of Columbia; United States
DT Article
FS U.S. Imprints not USDA, Experiment or Extension
LA English

L7 ANSWER 11 OF 11 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AB The oocyst wall is one of the components that permits cryptosporidia both the survive in the environment and to retain infectivity. With the aim of identifying Cryptosporidium proteins specifically expressed at the oocyst stage, we screened lambda-gt11 genomic libraries of **Cryptosporidium parvum** with both an oocyst antiserum and specific genetic probe. We isolated, from distinct libraries, two overlapping clones containing an open reading frame encoding a 1,252-amino-acid polypeptide. the analysis of the dedduced amino acid sequence revealed unusually high contents of cysteine, proline, and histidine. The sequence was also characterized by two distinct amino acid motifs, each repeated several times. The DNA sequences coding for the

amino acids repeats showed a high frequency of synonymous mutations, a result suggesting that the repeated motifs may be functionally and/or structurally important to the parasite. Antisera and monoclonal antibodies developed against a recombinant polypeptide encompassing the first 786 amino acids revealed that the corresponding protein in C. parvum had an apparent molecular weight of 190,000. Moreover, confocal microscopy analysis with immunofluorescence indicated that the protein was localized on the oocyst wall as a uniform stain and within the oocyst itself as bright granules in close association with the residual body.

AN 1993:388852 BIOSIS

DN PREV199396064152

TI Characterization and immunolocalization of a Cryptosporidium protein containing repeated amino acid motifs.

AU Ranucci, Lorella; Mueller, Hans-Michael; La Rosa, Giuseppe; Reckmann, Ingeborg; Gomez Morales, Maria Angeles; Spano, Furio; Pozio, Edoardo; Crisanti, Andrea (1)

CS (1) Ist. Parassitol., Univ. di Roma La Sapienza, P.le Aldo Moro 5, 00185 Rome Italy

SO Infection and Immunity, (1993) Vol. 61, No. 6, pp. 2347-2356. ISSN: 0019-9567.

DT Article

LA English

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